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The comparison of biomarkers released by hydropyrolysis and Soxhlet extraction from source rocks of different maturities

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The Permian Dalong Formation (P_2d) source rocks from the mature Guangyuan outcrop section and the overmature Wangcang outcrop section in Sichuan Basin were selected. The Soxhlet extraction and kerogen catalytic HyPy were conducted on the P_2d source rocks. The biomarkers obtained by both methods were compared to discuss their difference and the influence extent of thermal maturation on covalently bound biomarkers. The results show that covalently bound biomarkers can hardly be correlated to the corresponding free biomarkers due to severe thermal alterations and/or interference of migrated hydrocarbons in both outcrop sections and thermal maturation have much lower influence on covalently bound biomarkers in kerogens than on free biomarkers in Soxhlet extracts. The application of HyPy in study of biomarker geochemistry can reduce the thermal maturation effect on biomarkers ($R_o \leqslant 2.4\%$) to a greater extent and the interference of migrated hydrocarbons between interbedded layers. The covalently bound biomarkers released by HyPy are useful in the study of biomarker geochemistry and oil-source correlation on high-overmature source rocks.

hydropyrolysis, thermal maturation, Sichuan Basin, Dalong Formation

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Biomarkers are organic compounds derived from biomass and extensively exist in sediments and sedimentary rocks [1,2]. These organic molecules, especially steranes and terpanes, with complex structure and biogenetic indication, have been widely applied in the study of petroleum geochemistry. In the process of hydrocarbon generation and expulsion, the expelled oil can inherit the biomarker characteristics of the source rocks by retaining the carbon skeleton of geo-macromolecule [3]. Oil-source correlations are based on the concept that biomarker parameters can indicate source and depositional environment and reflect thermal evolution of organic matters.

Most of the marine source rocks in South China are at high-overmature stage [4,5]. The biomarkers obtained by Soxhlet extraction from high-overmature source rocks or bitumens are too low to satisfy the requirement of instrumental analysis [4,6–8], and are easily polluted by recent sediments and migrated hydrocarbons to discount their reliability. Meanwhile, the source-related biomarker parameters extracted from high-overmature source rocks would become a similar developing pattern and can not represent the original characteristic of organic matters [4]. Furthermore, it is difficult to get enough biomarkers by simple pyrolysis or chemical degradation from high-overmature source rocks due to their being hydrogen poor [8]. Thus it is problematic to get molecular geochemical information from high-overmature source rocks and to correlate them with potential oil reservoir by routine methods.

Fixed-bed Hydropyrolysis (HyPy) refers to an open-system pyrolysis at high hydrogen pressures (>10 MPa) in the presence of a dispersed sulphided molybdenum catalyst. Previous studies [8–15] suggested that HyPy can release

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higher yield of extractable organic matter (EOM) than Soxhelt extraction at various maturities. Love et al. [9] implied HyPy can maximize the yields of covalently bound aliphatic biomarkers without adversely affecting their stereochemistries from low mature kerogen. Murray et al. [12] suggested that the high hydrogen pressure used in HyPy appears to suppress the potential ability of clay minerals to promote cracking and isomerisation during pyrolysis. Some other studies [13,16–18] further suggested that the covalently bound biomarkers released by HyPy from kerogen usually show characteristics of low maturity. Liao et al. [18] used HyPy to release covalently bound biomarkers from artificial thermal altered bitumens of various maturities (easy R_o: 1.08%–2.86%). The results by Liao et al. [18] indicated that those biomarkers are much more insensitive to thermal alteration than free biomarkers. HyPy are also used to release covalently bound biomarkers from high-overmature source rocks. For example, Sun et al. [8] used covalently bound biomarkers released by HyPy to study the palaeoenvironment of source rocks from the Tarim Basin. Therefore, HyPy seems to be a promising method to recover original geochemical information from high-overmature source rocks. However, some basic mechanisms concerning high-overmature source rocks need to be elucidated. First, whether the covalently bound biomarkers released by HyPy can be directly compared with the biomarkers obtained by Soxhlet extraction at high-over maturity stage? Second, whether the availability of the covalently bound biomarkers from kerogens used in source characterization and oil-source correlation can be influenced by thermal maturation?

In the Sichuan Basin, the Permian Dalong Formation (P_2d) source rocks comprise marine dark silicalites and mudstones that are interbedded [19–22], and mainly occur in the Guangyuan-Wangcang trough and the Western Hubei-Chengkou trough [19–21,23,24]. Most of the P_2d source rocks in the Sichuan Basin (such as in the Wangcang outcrop section) are at high-overmature stage, with very low amount of EOM. However, the P_2d source rocks in the Guangyuan outcrop section are still in oil-generative window due to the uplift of Longmen Mountain. The P_2d source rocks in the Wangcang and the Guangyuan outcrop sections shared very similar depositional environments, since they occur in the same trough. Therefore, the P_2d source rocks from the Guangyuan and the Wangcang outcrop sections were selected for Soxhlet extraction and HyPy. The characteristics of the biomarkers released by HyPy were compared with those by Soxhlet extraction and the influences of thermal maturation on covalently bound biomarkers were discussed.

1 Samples and experiments

1.1 Source rock samples

The Upper-Permian Dalong Formation (P_2d) source rocks from the Guangyuan (GY series) and the Wangcang (WC series) outcrop sections shared very similar depositional environment but with different maturity, in the Sichuan Basin, China, were selected for this study (Figure 1). The P_2d Formation in the Sichuan Basin are typically marine dark

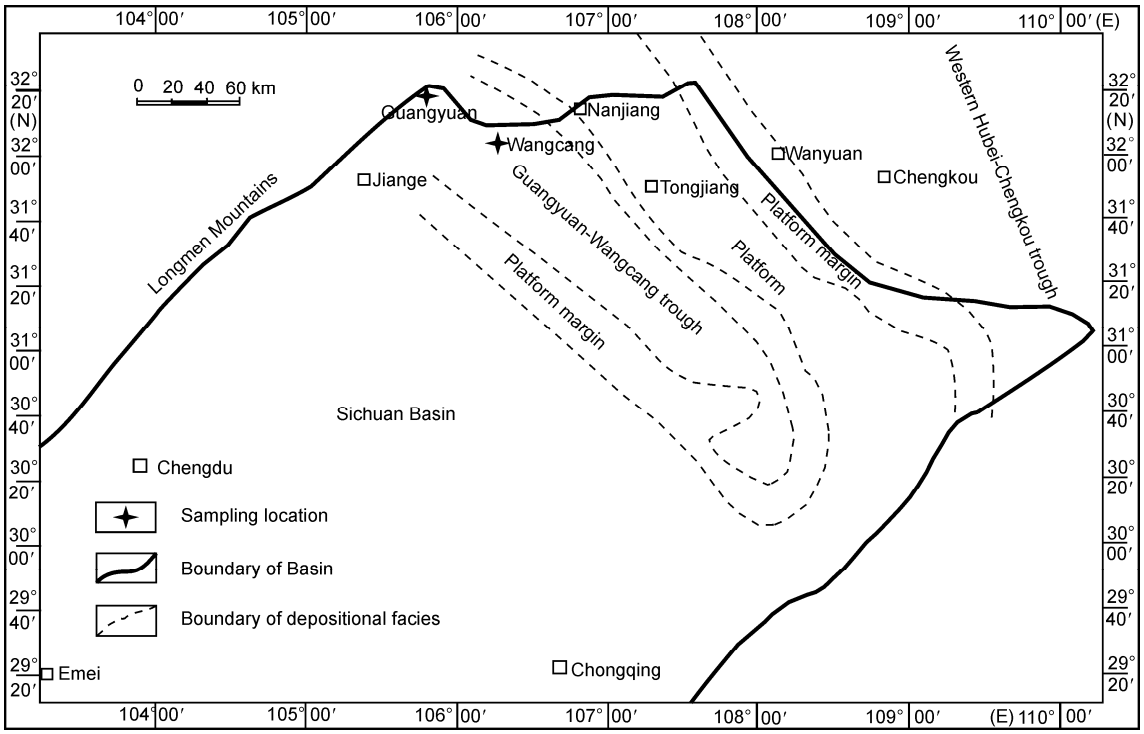

Figure 1 The geological map showing sampling location.

silicalite layers interbedded with mudstone layers [19,20]. Silicalites and mudstones are thought to be deposited in different depositional environments in that silicalites had deeper depositional water-depth than mudstones [25]. All the P_2d source rocks were sampled chronologically from old to young strata on each outcrop section, with intervals from 0.2 m to 0.5 m. The basic information of the samples is shown in Table 1. The T_{max} values of the GY series samples range from 436 to 438°C, with measured vitrinite reflectance (R_o) about 0.6%, implying that the GY series samples are still in oil-generative window. The WC series samples are overmature, with the T_{max} values ranging from 552 to 603°C. R_o values of the samples from the Permian section of the Wangcang area measured by Liang et al.[1)] are in the range of 1.88%–2.39%. Micrinites are dominant macerals in the kerogens of the P_2d source rocks. Vitrinite can be occasionally found in the GY series kerogens but at low concentration. Meanwhile, their bulk $\delta^{13}C$ are in very narrow range of −27.1‰– −28.4‰. The kerogens in the P_2d source rocks are of type II in the study area.

1.2 Dry Pyrolysis and HyPy experiment

The dry pyrolysis experiments were conducted on the kerogen GY-8. The kerogen was sealed in glass tubes under the protection of nitrogen gas. The tubes were then heated for 72 h at 400, 430 and 460°C, respectively. The calculated vitrinite reflectance (R_o) by Easy $\%R_o$ method [26] for the kerogen heated at 400, 430 and 460°C were 1.74%, 2.27% and 2.86%, respectively (the maturities of P_2d samples from the WC series are in this range). The pyrolysates were recovered by adding dichloromethane (DCM), and then sonicated and washed repeatedly.

The source rock samples were crushed to 80 mesh and then extracted with a mixture of DCM and methanol (93:7, v/v) at 46°C for 72 h. Then, those rock powders were treated by acid to get kerogens. The kerogen samples (0.3–0.8 g) for HyPy were Soxhlet extracted by ternary solvent (benzene/acetone/methanol=5:5:2) for 2 weeks to remove soluble organic matter. HyPy apparatus were described by Love et al. [9]. The solvent-extracted kerogens were impregnated with an aqueous solution of ammonium dioxydithiomolybdate [$(NH_4)_2MoO_2S_2$] to give a nominal loading of molybdenum of 5 wt%. Then, the kerogen powders with catalyst were loaded into stainless steel reactor to do hydropyrolysis using a hydrogen pressure of 15.0 MPa. A hydrogen flow of 4 L/min, measured at ambient temperature and pressure, was used through the reactor. The kerogen powder was then vacuum-dried for subsequent HyPy experiment. HyPy experiment involves two steps, both steps were conducted in the same hydrogen pressure and hydrogen flow. At first, the sample was pyrolysed with resistive heating from ambient temperature programmed to 300°C (5 min) at 250°C/min rate, to completely remove the adsorbed hydrocarbons and the weaker covalent bonds leaving the stronger bonds for HyPy [9]. After all of the weaker covalently bound compounds were removed from the kerogen, the collecting tube was replaced. The second heating run was employed from ambient temperature programmed to 250°C at 300°C/min rate, then increase to 520°C at 8°C/min rate, and finally kept at 520°C for 5 min. Before each HyPy experiment, the collecting tube of HyPy system was washed with DCM and then heated at 600°C for 4 h. A HyPy blank experiment was performed before running real samples. The details of HyPy experiments can be referred to Sun et al. [8] and Liao et al. [18]. The hydropyrolysates were collected by clean gel silica in a liquid nitrogen cold trap and recovered by DCM/methanol (93:7, v/v) for subsequent fractionation and analysis.

The pyrolysates and the Soxhlet extracts as well as the hydropyrolysates of the P_2d source rocks were all conducted on the following process. The details of Soxhlet extraction can be referred to Zhang et al. [27] and Liao et al. [28]. The asphaltenes were precipitated from the extracts by adding 50:1 (v/v) cold *n*-hexane, and then removed by centrifugation. The maltene fractions were fractionated by silica/alumina (3:1, v/v) column chromatography into saturate, aromatic and polar fractions, which were eluted with

Table 1 The basic information of the source rocks

Sample ID	Lithology	$\delta^{13}C$ (‰)	R_o (%)	T_{OC} (%)	*S1*	*S2*	T_{max} (°C)	HI
GY-3	mudstone	−28.4	−	3.54	0.43	7.29	437	206
GY-8	silicalite	−27.6	0.58	8.75	1.22	30.06	438	343
GY-9	siliceous mudstone	−27.1	−	4.58	0.48	12.68	436	300
GY-17	mudstone	−27.8	0.68	3.67	0.61	13.31	438	357
WC-4A	silicalite	−28.1	−	2.81	0	0.1	603	5
WC-5	mudstone	−27.5	−	4.07	0.04	0.18	601	5
WC-6A	silicalite	−27.1	−	8.54	0.01	0.15	552	2
WC-6B	mudstone	−27.3	−	7.10	0	0.15	556	2

1) Liang D G, Chen J P, Zhao Z, et al. Evaluation on efficient source rock of complex structure area in Southern China (unpublished report, in Chinese). SINOPEC Exploration Southern Company, 2007

n-hexane, DCM/*n*-hexane (3:1, v/v) and DCM/methanol (2:1, v/v), respectively. The saturated hydrocarbon fractions were analyzed by GC-MS. The saturated hydrocarbon fraction resulting from HyPy blank experiment and subsequent column chromatography is also analyzed by GC-MS. There are no steranes and terpanes identified (Figure 2).

1.3 Instrumental

Analysis of the saturated biomarkers was conducted using a Thermo Scientific Trace GC Ultra gas chromatography coupled to a Thermo Scientific Trace DSQ II mass spectrometer. A DB-1 fused silica capillary column (60 m × 0.32 mm i.d. × 0.25 μm film thickness) was used. The GC oven was held isothermally at 70°C for 2 min, programmed to 290°C at 4°C/min rate, with a final holding time of 30 min. Helium was used as carrier gas with a constant flow rate of 1.5 mL/min. The ion-source temperature was 250°C, and the temperature of injector was 290°C. The ion source was operated in the electron impact (EI) mode with electron energy of 70 eV.

2 Results and discussion

In both the Guangyuan outcrop section (GY series) and the Wangcang outcrop section (WC series), the TOC normalized yields of Soxhlet extraction and HyPy and their saturates/aromatics ratios are listed in Table 2. The TOC normalized EOM yields by Soxhlet extraction from the WC series are in the range of 0.7–1.2 mg/g TOC, much lower than those from the GY series (45.4–92.6 mg/g TOC) due to the high maturity. The TOC normalized EOM yields by HyPy are higher than those by Soxhlet extraction for both outcrop sections. The TOC normalized EOM yields by HyPy are 4–9 times those by Soxhlet extraction in the GY series. However, the TOC normalized EOM yields by HyPy are 37–168 times those by Soxhlet extraction in the WC series. In hydropyrolysates, saturated hydrocarbon fractions make 9.4%–27.9% of total EOM for the GY series, contrast to 0.2%–2.7% for the WC series. It indicates that the kerogen is high in aromaticity and there are few aliphatic structures in the kerogen structure at high-overmature stages due to severe decomposition and condensation.

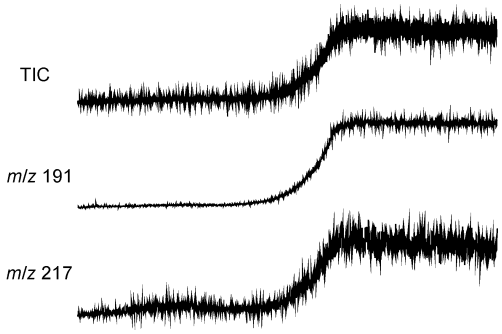

Figure 2 TIC, *m/z* 191 and *m/z* 217 mass chromatograms of HyPy blank.

2.1 Source-related biomarker parameters

The steranes are considered to be sourced from sterols in eukaryotic organisms [3,29]. The sterane ternary diagram of $C_{27–29}$ ααα20R is commonly used to distinguish groups of crude oils from different source rocks or different organic facies of the same source rock [3,30,31], and also extensively used to show relationships between oils and/or source rocks [32,33].

There are significant differences in the distribution of $C_{27–29}$ ααα20R steranes between the GY and the WC series. The distribution of C_{27-29} ααα20R steranes in all P_2d source rock extracts from the GY series show similar "V" shape ($C_{27} \approx C_{29} > C_{28}$), which was also reported by Tenger et al. [21]. However, the distribution of $C_{27–29}$ ααα20R steranes in P_2d source rock extracts from the WC series show similar "L" shape ($C_{27} > C_{29} > C_{28}$) (Figure 3). Study by Lu et al. [34] suggested that secondary cracking in thermal maturation may alter the distribution of $C_{27–29}$ ααα20R steranes because C_{29} and/or C_{28} ααα20R steranes show lower thermal stability than C_{27} ααα20R steranes. Thus the sample GY-8 (R_o: 0.58%) was selected for artificial thermal maturation simulations to investigate the influence of thermal maturation on the distribution of $C_{27–29}$ ααα20R steranes in the Soxhlet

Table 2 The TOC normalized yields of Soxhlet extraction and HyPy

Sample ID	Soxhlet extracts (mg/g TOC)			Hydropyrolysates (520°C, mg/g TOC)			Hydropyrolysate/ Soxhlet extract
	EOM (mg/g TOC)	Saturates (%)	Saturates/aromatics	EOM (mg/g TOC)	Saturates (%)	Saturates/aromatics	
GY-3	92.4	12.9	0.85	374.6	23.9	0.66	4
GY-8	56.0	7.3	0.24	497.7	22.7	0.66	9
GY-9	82.7	11.2	0.28	472.7	27.9	0.96	6
GY-17	45.4	18.3	0.40	205.0	9.4	0.12	5
WC-4A	0.7	14.3	0.25	32.9	2.7	0.05	47
WC-5	1.2	8.3	0.17	44.7	1.3	0.03	37
WC-6A	0.7	14.3	1.00	117.3	0.2	0.01	168
WC-6B	1.0	10.0	0.14	146.4	0.5	0.01	146

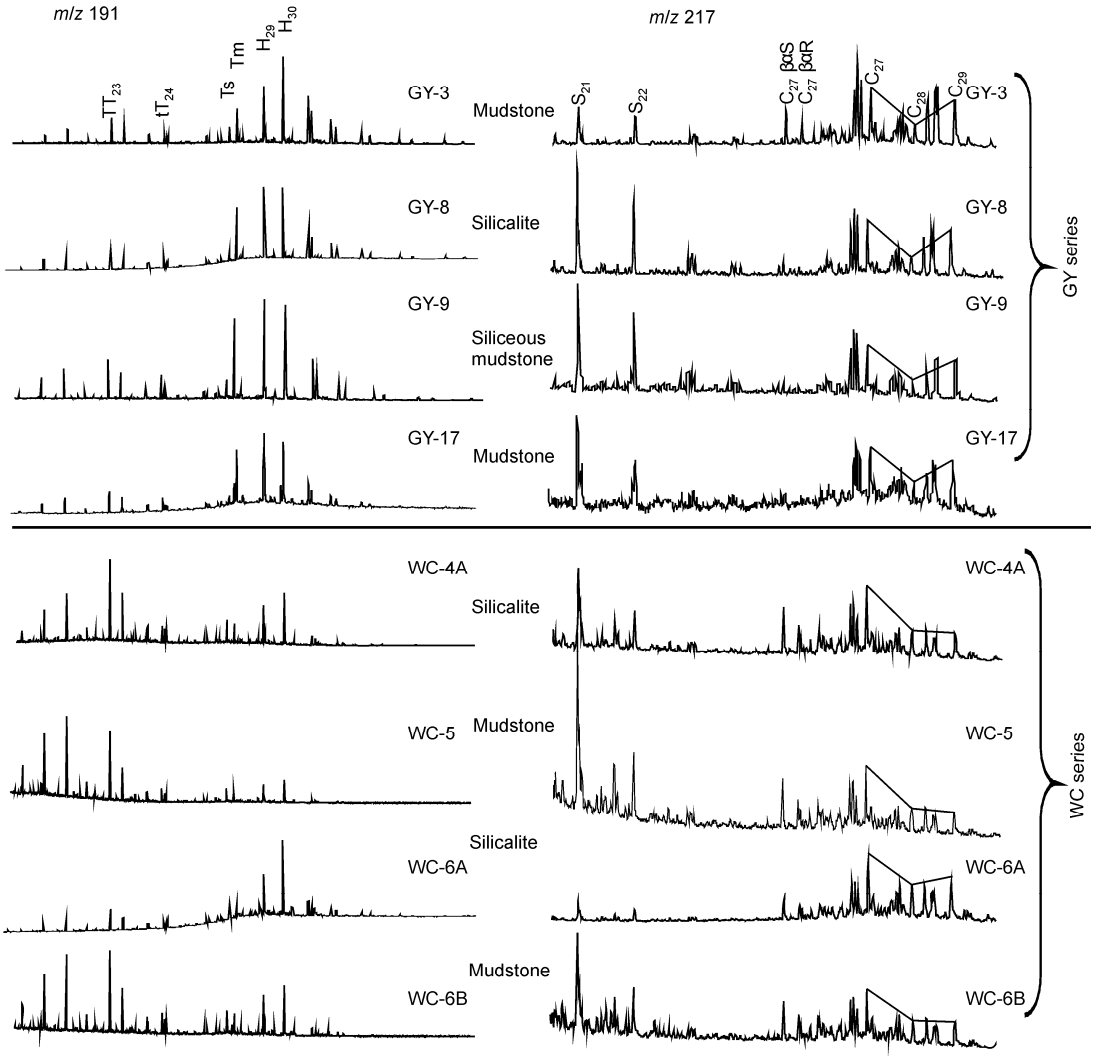

Figure 3 *m/z* 191 and *m/z* 217 mass chromatograms of the Soxhlet extracts from the Dalong Formation source rocks.

extract of P_2d source rock. The results show that the distribution of C_{27-29} ααα20R steranes change gradually from "V" shape in original extracts to "L" shape in the 72 h artificial thermal maturation products at 400–460°C (Figure 4) due to a quicker decrease in C_{29} sterane (Figure 5(a)). Such variations resulting from increasing maturity can also be clearly seen in the sterane ternary diagram of C_{27-29} ααα20R in Soxhlet extracts from both outcrop sections. Namely, the abundance of C_{29} sterane relative to C_{27} sterane in the Soxhlet extracts of the GY series are much higher than that of the WC series (Figure 5(b)). Therefore, the difference in the distribution of C_{27-29} ααα20R steranes in the Soxhlet extracts between the GY and the WC series can be attributed to the difference in maturity. It indicated that the source-related biomarker parameters in Soxhlet extracts of high-overmature source rocks can be altered by thermal maturation and the original characteristic of organic matters may also be altered.

The biomarker assignments are listed in Table 3.

The P_2d source rocks in the Sichuan Basin are typically marine dark silicalite layers interbedded with mudstone layers. In hydropyrolysates, the distribution of C_{27-29}ααα20R steranes in the silicalite layers have a C_{29} advantage, while

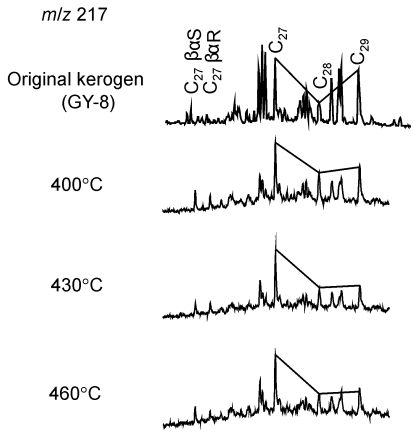

Figure 4 *m/z* 217 mass chromatograms of the Soxhlet extracts of original sample and the pyrolysates of artificial simulation series.

those in the mudstones layer show a C_{27} advantage (Figure 6). The hydropyrolysates from mudstone layers and that from silicalite layers are in two different groups in the sterane ternary diagram (Figure 5(c)). Interestingly, the hydropyrolysates of the siliceous mudstone layer sample GY-8 is in between the two groups (Figure 5(c)). The hydropyrolysates from the samples with the same lithology have similar distribution of C_{27-29} ααα20R (Figure 5(c)) in spite of different maturities. It seems that the depositional environment should be the major factor controlling the distribution of the covaltently bound C_{27-29} ααα20R steranes (Figures 5(c) and 6). The distribution of covalently bound steranes released by HyPy from high-overmature source rocks are similar to those from the mature source rocks for the protection of macromolecular structure [17,35]. Therefore, the covalently bound steranes released by HyPy from the overmature Permian Dalong Formation source rocks in the WC series (R_o≤2.4%) can still represent the original characteristic

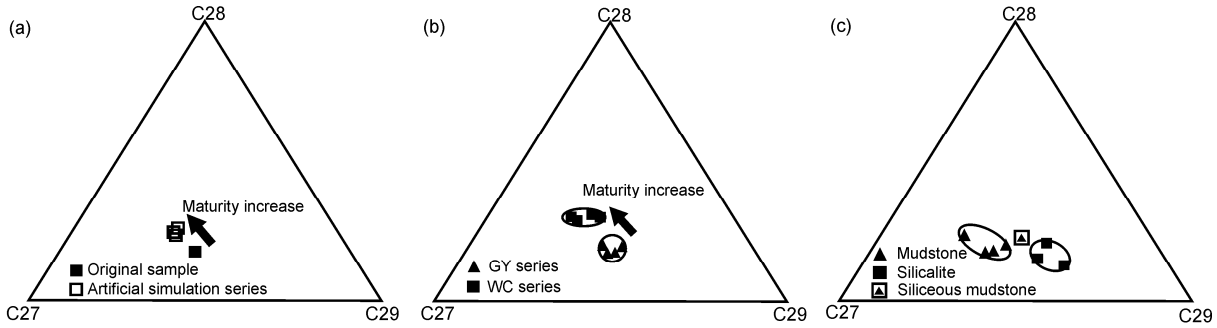

Figure 5 The ternary plot of C_{27-29} ααα20R sterane distribution. (a) Soxhlet extracts of GY-8 and artificial thermal maturation products; (b) Soxhlet extracts. (c) Hydropyrolysates.

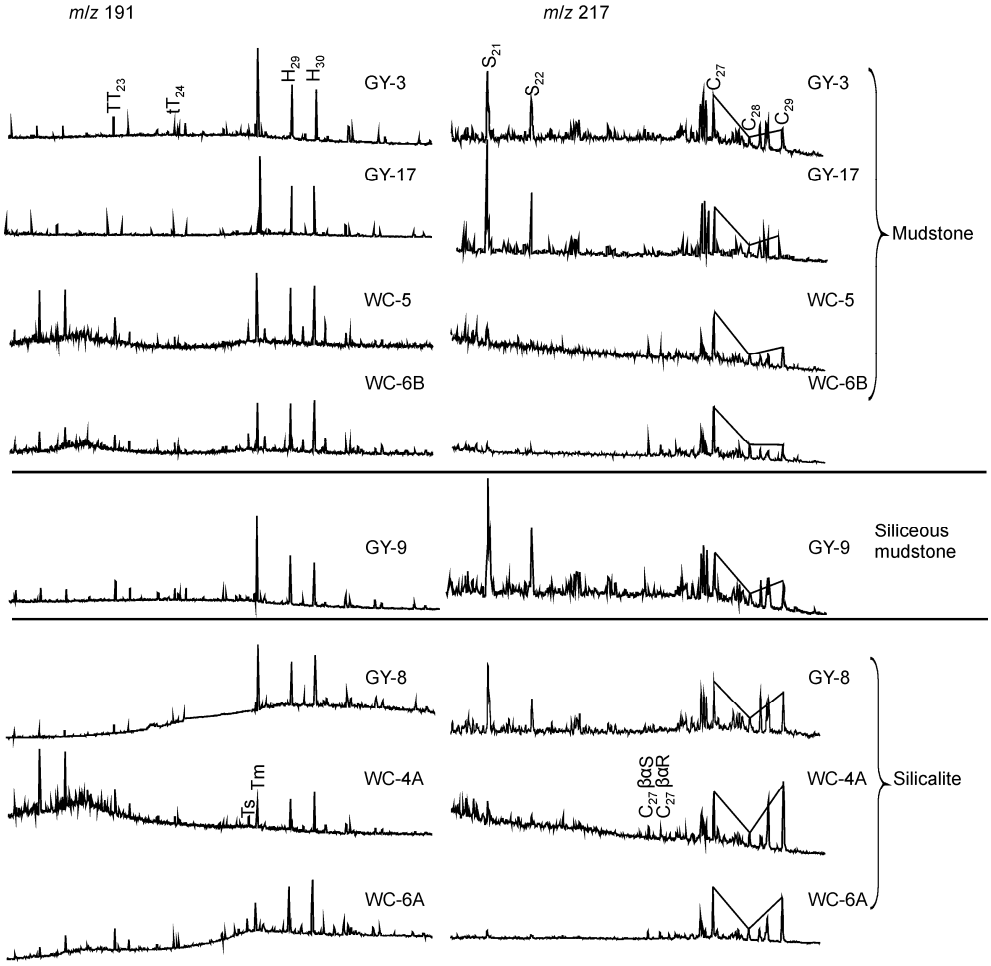

Figure 6 *m/z* 191 and *m/z* 217 mass chromatograms of the hydropyrolysates from P_2d source rocks.

Table 3 Biomarker assignments

Peak	Compound
TT_{23}	C_{23}-tricyclic terpane
tT_{24}	C_{24}-tetracylic terpane
H_{29}	C_{29}-αβ hopane
H_{30}	C_{30}-αβ hopane
Ts	C_{27} 17α-trisnorhopane
Tm	C_{27} 18α-trisnorhopane
C_{27}	C_{27}-ααα R cholestane
C_{28}	C_{28}-ααα R ergostane
C_{29}	C_{29}-ααα R stigmastane
C_{27} βαS	C_{27} βαS diacholestane
C_{27} βαR	C_{27} βαR diacholestane
S_{21}	C_{21} Pregnane
S_{22}	C_{22} Homopregnane

of organic matter.

As discussed above, there are differences in the distribution of covalently bound C_{27-29} ααα20R steranes between silicalite layers and mudstone layers from the GY series (Figure 6). However, in the Soxhlet extracts of both silicalite layers and mudstone layers from the GY series, C_{27} ααα20R sterane shows similar abundance with C_{29} ααα20R sterane ($C_{27} \approx C_{29} > C_{28}$, Figure 3). Actually, this should be attributed to the mixing of migrated hydrocarbons of C_{27} advantage from mudstone layers with migrated hydrocarbons of C_{29} advantage from interbedded silicalite layers. It is also supported by field work observation. In the Guangyuan outcrop section, expelled hydrocarbons can be observed in fissures of the silicalite layers. Those migrated hydrocarbons can easily enter the interbedded layers through pores and fissures. Therefore, the biomarker characteristics of P_2d source rock extracts in the GY series actually reflect a kind of mixing between the hydrocarbons from the silicalite layers and interbedded mudstone layers. However, the covalently bound biomarkers in kerogens were not influenced by migrated hydrocarbons because migrated hydrocarbons can hardly incorporate into the kerogen macromolecular structures. Additionally, kerogen powders were extracted by ternary solvent for 2 weeks to sufficiently remove adsorbed hydrocarbons before HyPy experiment. Therefore, the HyPy technology can reduce the interference of migrated hydrocarbons on high-overmature source rocks, and the covalently bound biomarkers released from high-overmature source rocks by HyPy can represent the original characteristics of original organic matter.

The major precursors to pregnane and homopregnane were considered to be hormones, pregnanol and pregnanone [36]. Huang et al. [37] thought that pregnanes and homopregnanes were sourced partly from the precursors in kerogen, and partly from thermal cracking of C_{27-29} regular steranes. Huang et al. [37] also pointed out that the relative abundance of pregnanes to regular steranes is low at low mature stage (R_o<0.42%), and sharply increase at mature stage (R_o>0.42%). Both in the GY and the WC series, the Soxhlet extracts have high abundance of pregnane and homopregnane relative to regular steranes (Figures 3 and 6), because the maturities of source rocks from both outcrop sections are high enough to generate abundant hydrocarbons [37]. In hydropyrolysates, the relative abundances of pregnane to regular steranes are also high in the GY series, but low in the WC series. Large mounts of hydrocarbons were released from kerogen with increasing maturities, which resulted in significant decrease in the covalently bound pregnanes. Therefore the covalently bound pregnanes has almost been removed completely from kerogens of the WC series samples by thermal maturation. However, the GY series samples are still in oil generative window with a great number of pregnanes still incorporated in kerogen. So the hydropyrolysates of the GY series samples still have very high relative abundance of pregnanes. The pregnane/homopregnane (S_{21}/S_{22}) ratio in extracts of source rocks from both outcrop sections range from 1.05 to 2.46, while S_{21}/S_{22} ratio in hydropyrolysates are in the range of 1.42–1.78, more stable than that in the corresponding Soxhlet extracts. The results suggested that the relative abundance of pregnane and homopregnane released by HyPy decrease with increasing maturity, but the ratio of S_{21}/S_{22} is still quite stable.

The ratio of C_{23}-tricyclic terpane to C_{23}- and C_{24}-tricyclic terpanes ($TT_{23}/(TT_{23}+TT_{24})$) remains constant in the Soxhlet extracts and hydropyrolysates (Figures 3, 4, Table 4), indicating that tricyclic terpanes have high resistance to both biodegradation and thermal alteration than pentacyclic terpanes. The ratio of C_{23}-tricyclic terpane to C_{30}-hopane (TT_{23}/H_{30}) is commonly used as source-related parameters in oil generative window [3]. In Soxhlet extracts, the ratio TT_{23}/H_{30} of the WC series (0.21–2.18) is higher than that of the GY series (0.11–0.28). Meanwhile, the TT_{23}/H_{30} ratio in the hydropyrolysates of the WC series ranges from 0.12 to 0.52, more stable and lower than that in the corresponding Soxhlet extracts, since tricyclic terpanes are more resistant to thermal degradation than hopanes [3]. The ratio of C_{29}-norhopane to C_{30}-hopane ratios (H_{29}/H_{30}) is also a commonly used source-related biomarker parameter [3]. H_{29}/H_{30} ratio varied in the range of 0.50–1.08 in the Soxhlet extracts from both outcrop sections. However, H_{29}/H_{30} ratio in the hydropyrolysates ranges from 0.76 to 0.93 in mudstones and from 0.67 to 0.73 in silicalites. H_{29}/H_{30} in hydropyrolysates seems more stable than in the corresponding Soxhlet extracts. It implies that the covalently bound biomarkers are not significantly altered by thermal maturation when $R_o \leqslant 2.4\%$ due to the protection of macromolecular structure [17,35].

In conclusion, though the source-related biomarker parameters such as the distribution of C_{27-29} ααα20R steranes, the ratios of TT_{23}/H_{30} and H_{29}/H_{30} in Soxhlet extracts were significantly altered by thermal maturation at high-over-

Table 4 The biomarker parameters of the Soxhlet extracts and hydropyrolysates of P_2d source rocks

Biomarker parameters	Soxhlet extracts								Hydropyrolysates							
	Mudstone				Siliceous Mudstone	Silicalite			Mudstone				Siliceous Mudstone	Silicalite		
	GY-3	GY-17	WC-5	WC-6B	GY-9	GY-8	WC-4A	WC-6A	GY-3	GY-17	WC-5	WC-6B	GY-9	GY-8	WC-4A	WC-6A
$C_{27}/(C_{27}+C_{28}+C_{29})$	37	43	45	39	40	43	46	36	45	52	51	56	41	34	33	40
$C_{28}/(C_{27}+C_{28}+C_{29})$	19	17	29	31	17	20	30	31	19	16	17	24	21	21	12	14
$C_{29}/(C_{27}+C_{28}+C_{29})$	43	41	26	30	42	38	24	32	36	32	32	21	39	45	55	46
S_{21}/S_{22}	1.05	1.40	2.46	1.89	1.19	1.20	1.85	1.44	1.51	1.78	1.75	1.63	1.45	1.42	1.69	1.70
C_{27}-βαR/C_{27}ααR	0.38	–	0.56	0.57	0.15	0.08	0.44	0.26	–	0.13	0.19	0.22	–	–	0.19	0.12
C_{29}S-ββ/(ββ+αα)	0.56	0.46	0.41	0.40	0.52	0.57	0.40	0.39	0.51	0.48	0.35	0.39	0.48	0.42	0.40	0.34
C_{29}-20S/(20S+20R)	0.49	0.43	0.53	0.52	0.44	0.45	0.57	0.51	0.49	0.47	0.28	0.41	0.52	0.50	0.14	0.20
Ts/(Ts+Tm)	0.32	0.14	0.51	0.50	0.17	0.17	0.51	0.53	0.06	0.06	0.17	0.26	0.04	0.05	0.27	0.27
$TT_{23}/(TT_{23}+TT_{24})$	0.57	0.60	0.69	0.66	0.60	0.59	0.64	0.63	0.67	0.60	0.66	0.64	0.66	0.64	0.69	0.68
TT_{23}/H_{30}	0.21	0.15	2.18	1.16	0.28	0.11	1.19	0.21	0.25	0.27	0.33	0.21	0.17	0.12	0.52	0.27
H_{29}/H_{30}	0.61	1.08	0.70	0.67	0.91	0.86	0.66	0.50	0.89	0.93	0.89	0.76	0.88	0.73	0.68	0.67
H_{31}-22S/(22S+22R)	0.61	0.58	0.60	0.60	0.62	0.60	0.60	0.58	0.60	0.58	0.52	0.53	0.58	0.60	0.60	0.56

mature stages, they were quite stable in the hydropyrolysates. The reason is that the covalently bound biomarkers released by HyPy from kerogen [9,35] and/or bitumen [18] were protected by the macromolecular structure from secondary alterations such as thermal maturation [17,18,35] and biodegradation [38–41]. Additionally, the covalently bound biomarkers released by HyPy from kerogen can eliminate the interference of migrated hydrocarbons between interbedded layers and retain original organic geochemical information.

2.2 Maturity-related biomarker parameters

Diasteranes/steranes ratios are commonly used to distinguish petroleum from carbonates or clastic source rocks [3,42]. However, high diasteranes/steranes ratios can also result from high thermal maturity [43] and/or heavy biodegradation [44,45]. Thus the ratio of C_{27} cholestane to C_{27} diacholestane (C_{27}-βαR/C_{27}-ααR) was calculated and compared (Table 4). In Soxhlet extracts, the ratios C_{27}-βαR/C_{27}-ααR in the source rocks from the WC series are higher than that from the GY series, which may be attributed to the maturity differences of the source rocks in the two outcrop sections. Meanwhile, the ratios C_{27}-βαR/C_{27}-ααR in the hydropyrolysates are lower than that in the corresponding Soxhlet extracts, implying that the covalently bound biomarkers released by HyPy have lower maturity than the free biomarkers.

Most isomerization ratios related to maturities in free biomarkers obtained by Soxhlet extraction are only valid in the oil generative window or at even lower maturity stage because these ratios reach their end point at high maturities. The C_{29} sterane isomer ratios of C_{29}-ββ/(ββ+αα) and C_{29}-20S/(20S+20R) are commonly used as maturity-related indicators in oil-generative window [3]. The isomerization at C-14 and C-17 in the C_{29} regular steranes causes an increase in C_{29}-ββ/(ββ+αα) from 0 to ca. 0.7 (0.67–0.71 end point) with increasing thermal maturity. The C_{29}-20S/(20S+20R) ratio rises from 0 to ca. 0.5 (0.52–0.55 end point) with increasing isomerization at C-20 during thermal maturation [3,31]. The sterane isomerization ratio C_{29}-ββ/(ββ+αα) in the hydropyrolysates of the P_2d source rocks from the GY series are in the range of 0.34–0.40 (Table 4), while in the corresponding Soxhlet extracts it is in the range of 0.39–0.41. Thus the ratio C_{29}-ββ/(ββ+αα) is in similar range in both hydropyrolysates and Soxhlet extracts. However, the ratio C_{29}-20S/(20S+20R) in the hydropyrolysates of the P_2d source rocks of the WC series is in the range of 0.14–0.41, but in the Soxhlet extracts it is in the range of 0.52–0.57. Thus for the P_2d source rocks of the WC series, the C_{29}-20S/(20S+20R) ratio in the hydropyrolysates is much lower than in the Soxhlet extracts. Such results were also reported in previous catalytic hydropyrolysis studies [12,13,16,17]. In the GY series, the C_{29}-ββ/(ββ+αα) ratio in the hydropyrolysates are in the rang of 0.42–0.51, while it is in the range of 0.46–0.57 in the corresponding Soxhlet extracts. The C_{29}-20S/(20S+20R) ratio in the hydropyrolysates are in the rang of 0.47–0.52, but in the Soxhlet extracts it is in the range of 0.43–0.49. Therefore, the C_{29}-ββ/(ββ+αα) and C_{29}-20S/(20S+20R) ratios in the hydropyrolysates are quite similar to their counterparts in the Soxhlet extracts for the GY series. However, in this study the covalently bound maturity-related biomarkers released by HyPy from kerogen in the overmature WC series show lower maturities than those in the mature GY series. Lockhart et al. [17] also reported that the isomerization ratios of both C_{29} steranes and C_{31} hopanes in hydropyrolysates of kerogen of high maturities are lower than those in hydropyrolysates of kerogen of lower matuirties in the T_{max} range of 407–464°C. It implied that at high maturity stage the influence of thermal maturation on the covalently bound biomarkers released by HyPy from kerogen may be different from that on the free counterparts in Soxhlet extracts, further study is needed to elucidate how such difference occur.

Isomerization of 17α-hopanes at C-22 occurs earlier than that of regular steranes at C-20 [3]. Hence, the 22S/(22S+22R) homohopane isomerization ratio for C_{31}–C_{35} hopanes is highly specific for immature to early oil generation stages. Table 4 shows that the isomerization ratios of C_{31}-hopane (H_{31}-22S/(22S+22R)) in Soxhlet extracts and hydropyrolysates are all about 0.6, very close to the end point. Tm (C_{27} 18α-trisnorhopane) is less stable than Ts (C_{27} 17α-trisnorhopane), and can be transformed to Ts with increasing maturity [43,46]. Ts/(Ts+Tm) ratio depends on both of source and maturity [47]. It is commonly used as maturity indicator of oils from source rocks with the same organic facies [3]. The Ts/(Ts+Tm) ratios in the Soxhlet extracts for the GY series range from 0.14 to 0.32. It reached its end points (ca. 0.5) in the WC series because of high maturity. There is only trace Ts in the hydropyrolysates for the GY series. It is also reported in previous studies [10,48] that Ts was absent in the hydropyrolysates of low mature kerogen. The Ts/(Ts+Tm) ratio of the hydropyrolysates is significantly lower than that of the Soxhlet extracts for the WC series. Previous studies [12,49–51] suggested that the carbon skeletons of bacteriohopanepolyols precursors are cross linked to kerogen geo-macromolecular networks by multiple heteroatomic linkages. Thus, Tm was restrained to be transformed into Ts in the hydropyrolysates because the covalently bound biomarkers were protected by geo-macromolecular networks. Furthermore, high openness of HyPy pyrolysis system also restricts the conversion of Tm to Ts by quickly blowing the hydropyrolysates away from the high temperature reactor tube with high pressure hydrogen.

The release of biomarkers from macromolecules (kerogen and bitumen) can lead to the decrease in isomerization ratios [13,18,34,52,53], while isomerization and decomposition (thermal maturation) can lead to the increase in isomerization ratios [54–56]. Previous studies [54–56] demonstrated that the increase in isomerization ratios of hopanes and steranes was mainly resulted from the decomposition of biomarkers rather than direct isomerization. The parameters of maturity-related biomarkers extracted from high-overmature source rocks have already reached their end point [4]. However, the covalently bound biomarkers released by HyPy from kerogen [9,35] and/or bitumen [18] were protected by the macromolecular structure from secondary alterations such as thermal maturation [17,18,35] and biodegradation [38–41]. Meanwhile, the decomposition and isomerization of the covalently bound biomarkers released by HyPy were suppressed because HyPy is a kind of open-system pyrolysis using high pressure hydrogen as carrier gas. Thus the covalently bound biomarkers released by HyPy from kerogen show lower maturity than their counterparts in Soxhlet extracts. Therefore, the covalently bound biomarkers released by HyPy from kerogen are less altered by thermal maturation than their counterparts in Soxhlet extracts.

3 Conclusions

The characteristics of biomarkers in hydropyrolysates and Soxhlet extracts from two Permian Dalong Formation outcrop sections, with similar depositional environments and very different maturities, in Guangyuan and Wangcang areas were compared and discussed. Based on these results, the following conclusions can be made:

(1) Considerable amount of covalently bound biomarkers released by HyPy from high-overmature Permian Dalong Formation source rock kerogen in the Wangcang outcrop section ($R_o \leqslant 2.4\%$) can meet the requirement of instrumental analysis. The yields of EOM by HyPy are 37–168 times those by Soxhlet extraction. In hydropyrolysates, saturated hydrocarbons fraction make 9.4%–27.9% of total EOM for GY series, contrasted to 0.2%–2.7% for the WC series. It implied that there are few aliphatic structures in the kerogen structure at high-overmature stages due to severe decomposition and condensation.

(2) Thermal maturation have much lower influence on the covalently bound biomarkers in kerogens than on free biomarkers in Soxhlet extracts. The source-related biomarker parameters such as the distribution of $C_{27–29}$ ααα20R steranes, the ratios of the TT_{23}/H_{30} and the H_{29}/H_{30} were quite stable in the hydropyrolysates of various maturities. Most of the maturity-related biomarker parameters such as the isomerization ratios of hopanes and steranes were also lower than their counterparts in Soxhlet extracts.

(3) The free biomarkers in Soxhlet extracts of high-over mature source rocks can hardly be correlated to the corresponding covalently bound biomarkers due to severe thermal alterations. The application of HyPy to study of biomarkers can reduce the thermal maturation effect on covalently bound biomarkers ($R_o \leqslant 2.4\%$) to a greater extent. The free biomarkers in Soxhlet extracts of source rocks that are interbedded with layers of different lithology can hardly be correlated to the corresponding covalently bound biomarkers due to interference of migrated hydrocarbons between interbedded layers. The application of HyPy to study of biomarkers also can reduce the interference of migrated hydrocarbons. The covalently biomarkers released by HyPy can represent the original organic geochemical information and are useful in the study of biomarker geochemistry and oil-source correlation on high-overmature source rocks, providing a new approach for the geochemistry study of high-overmature source rocks in South China.

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